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## ORIGINALPATENT

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# Process for the production of monosodium glutamate as a feed supplement

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The present invention relates to a process for the production of monosodium glutamate, which as a supplement to animal feeds improves their flavor and appearance.

Monosodium glutamate has been obtained previously by hydrolysis of plant proteins, by synthesis and by fermentation. Free glutamic acid is obtained in all these processes which is then converted into the monosodium salt. The racemate is obtained by synthesis and its separation is expensive and time consuming. Only the L-form is suitable as a food additive.

The L-glutamate can be better obtained from L-glutamic acid from fermentation processes; however, the isolation from the fermentation broth is difficult and consequently expensive in this case also.

Crystallized L-monosodium glutamate has therefore been expensive up to this time and could be used as a food additive to only a limited degree.

The object of the present invention is to produce monosodium glutamate in an economical manner so that it is feasible to use it as an additive in animal nutrition and in so doing the salt is obtained very quickly in one step without isolation of the free acid.

The present invention represents an improvement of the fermentation method.

The monosodium glutamate is obtained from the fermentation broth in the form of a concentrated solution which is equivalent to the crystalline substance. In one embodiment of the process, the fermentation mixture is dried and used as such.

In accordance with the invention, glutamic acid is produced by fermentation and it is converted into the monosodium salt by the introduction of sodium ions into the nutrient broth toward the end of the fermentation. The salt is then isolated as a concentrated solution or as a component of a dry powder.

The present process is applicable to numerous forms of glutamic acid fermentation. A glutamic acid-producing microorganism such as *Brevibacterium divaricatum* or *Micrococcus glutamicus* is grown in a medium containing carbohydrate, nitrogen, growth promoting substance, mineral salts and trace elements. The work is done at ambient temperature and given pH. Ammonium hydroxide is a very suitable nitrogen source and regulates the pH at the same time. Sodium hydroxide is added toward the end to form the salt without purifying or isolating the free glutamic acid. In so doing, it is possible to replace up to 50% of the ammonium hydroxide by sodium hydroxide.

The sodium hydroxide can also partially replace other bases and suppliers of nitrogen of traditional fermentation methods.

Other sodium ion suppliers such as sodium carbonate, sodium lactate, etc., may also be used in place of sodium hydroxide. Sodium hydroxide is preferred, however, since no undesirable byproducts are produced; in addition, it is easy to handle and inexpensive.

The L-monosodium glutamate preparation is isolated by evaporation. The clear, dark, viscous liquid can if necessary be filtered and/or decolorized. The preparation can be used like crystalline L-monosodium glutamate.

A slightly colored, powdered product can be obtained by processes like spray drying, etc. of the nutrient broth. It can be processed into tablets and pills for better dosing.

*Example I*

10 liters of the following nutrient broth were filled into a 25 L fermentation vessel:

| | | |
|---|---|---|
| Glucose | 1000 | g |
| Urea | 20 | g |
| $K_2HPO_4$ | 10 | g |
| $MgSO_4 \cdot 7H_2O$ | 5 | g |
| $FeSO_4$ | 40 | mg |
| Wheat bran extract* | 400 | ml |
| Water | | up to 10 liters total volume |

*obtained from 100 g bran by boiling for 30 minutes in 1000 ml water and filtering off the insoluble component

The pH of the medium was adjusted to 7.0 and sterilized by high temperature for ten minutes at a slight excess pressure. It was then inoculated with *Brevibacterium divaricatum* NRRL B-2312. The fermentation was carried out at 30° with stirring and aeration. During this time, the pH was held at 7 by the addition of a total amount of 300 ml 28-30% ammonia liquor. After this amount of ammonia was consumed, additional neutralization was carried out with 60% caustic soda solution. An additional amount of 255 ml caustic soda solution was required until the end of fermentation. 41 g of free L-glutamic acid was formed, equivalent to 47 g of the monosodium salt per liter of nutrient broth. A portion of the broth was filtered and evaporated to a concentration of 300 g L-monosodium glutamate

per liter. After identical evaporation to low volume, the unfiltered portion was spray dried to a slightly colored powder and partially processed into tablets.

*Example II*

Fermentation is done as in Example I; however, the quantity of ammonia is reduced to 275 ml of the 23-30% solution and the 60% caustic soda solution is increased to 280 ml at the same time. The yield of L-monosodium glutamate was for all practical purposes the same.

*Example III*

Fermentation is done as in Example I; however, the quantity of ammonia is increased to a 325 ml 28-30% solution and the quantity of 60% caustic soda solution is reduced to 230 ml. The yield of L-monosodium glutamate was for all practical purposes identical.

*Example IV*

10 liters of the following nutrient broth are filled into a 25 L fermentation vessel:

| | | |
|---|---|---|
| Glucose | 1000 | g |
| Urea | 80 | g |
| $K_2HPO_4$ | 10 | g |
| $MgSO_4 \cdot 7H_2O$ | 5 | g |
| $FeSO_4$ | 40 | mg |
| Wheat bran extract* | 400 | ml |
| Water | | up to 10 liters total volume |

*obtained from 100 g bran by boiling for 30 minutes in 1000 ml water and filtering off the insoluble component

The pH of the nutrient broth was brought to 7 and it was sterilized for 10 minutes by heating at a slight overpressure. The broth was then inoculated with a culture of the L-glutamic acid-producing strain

*Brevibacterium divaricatum* NRRL B-2311. It was fermented at 30° with stirring and aeration. Portions of 20 g urea each were added after 18, 24 and 30 hours of fermentation time. After that, the pH was maintained at 7 by addition of 60% caustic soda solution. An amount of 255 ml of alkaline solution was required for this purpose. The yield of L-monosodium glutamate is equal to that of Example I. A portion of the fermentation mixture was filtered and concentrated; the other unfiltered portion was spray dried to a light, weakly colored powder after evaporation to low volume and partially processed into tablets.

The process in accordance with the invention is also applicable to analogous fermentation methods with other glutamic acid-producing organisms.

The product in accordance with the invention improves the flavor of animal feeds so that they are accepted much better by the livestock. Given free choice, sheep consumed 117% more of a feed containing 0.5% of the spray dried product of Example I than identical feed free of additive.

Six sheep in one pen were offered two identical feed rations of which one contained a 0.5% by weight of the spray dried product of Example I. The additive product contained 50% monosodium glutamate. Every two days the feed containers were switched to eliminate the factor of a preferred feeding place. The following table shows the feed consumption:

*Test of feed consumption*

| Batch | Number of animals | Content of glutamate-containing dry product | Total feed consumed in kg | |
|---|---|---|---|---|
| | | 0% | 23.0 | |
| 1 | 6 | 0.5% | 51.0 | |
| | | 0% | 23.8 | 1.25 |
| 2 | 6 | 0.5% | 51.3 | 1.7 |
| Average | | 0% | 23.5 | |
| | | 0.5% | 51.0 | |

*Feed composition*

| | | |
|---|---|---|
| Ground corn cobs | 17.9 | kg |
| Ground maize | 12.2 | kg |
| Alfalfa meal | 9.0 | kg |
| Molasses (cane sugar) | 2.25 | kg |
| Soy bean meal, 44% | 3.2 | kg |
| Dicalcium phosphate | 259 | g |
| Lime | 113 | g |
| Iodized salt | 227 | g |
| Vitamin A supplement, 10 000/gm | 20 | g |
| Vitamin D supplement, 1500/gm | 15 | g |
| Trace mineral salts | 45 | g |

CLAIM I

Process for the production of monosodium glutamate as an animal feed supplement, characterized by the fact that an L-glutamic acid producing microorganism is grown in a nutrient medium and sodium ions are added during the course of fermentation

DEPENDENT CLAIMS

1. Process in accordance with Claim I, characterized by the fact that an L-glutamic acid-producing organism is grown.

2. Process in accordance with Claim I, characterized by the fact that sodium hydroxide is used as the supplier of sodium ions.

3. Process in accordance with Claim I, characterized by the fact that a pH of 6-9 is maintained during fermentation by initially adding ammonium hydroxide until L-glutamic acid is formed and then continuing to add sodium hydroxide and ammonium hydroxide until the end of fermentation.

4. Process in accordance with Claim I, characterized by the fact that the fermentation medium is filtered and a monosodium glutamate concentrate is produced from it by evaporation.

5. Process in accordance with Claim I, characterized by the fact that the fermentation medium thus obtained is evaporated to a dry monosodium glutamate concentrate.

6. Process in accordance with Claim I, characterized by the fact that *Brevibacterium divaricatum* NRRL B-2311 is used as the microorganism in an aqueous nutrient liquid containing assimalable carbohydrate and nitrogen, whereby L-glutamic acid is obtained at 10-37° and at a pH of 6 to 9 under submerged aerobic conditions, and whereby the pH of 6-9 is maintained by the addition of sodium hydroxide during the later course of the fermentation.

7. Process in accordance with Claim I and Dependent Claim 6, characterized by the fact that *Brevibacterium divaricatum* NRRL B-2312 is used as the microorganism.

8. Process in accordance with Claim 8, Dependent Claims 6 and 7, characterized by the fact that ammonium hydroxide is used as the source of nitrogen.

## CLAIM II

Feed supplement produced according to the process in accordance with Claim I, characterized by the fact that it contains the water soluble components of the glutamic acid-containing fermentation medium.

## DEPENDENT CLAIMS

9. Feed supplement in accordance with Claim II, characterized by the fact that it contains the water soluble components of the L-glutamic acid-containing fermentation medium.

10. Feed supplement in accordance with Claim II, characterized by the fact that it contains the water soluble and insoluble components of the L-glutamic acid-containing fermentation medium.

11. Feed supplement in accordance with Claim II and Dependent Claim 10, characterized by the fact that it is present in dry form.

12. Feed supplement in accordance with Claim II and Dependent Claim 10, characterized by the fact that it is present in tablet form.

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